LIDOCAINE INFUSIONS IN PANCREATIC CANCER: TRANSLATIONAL STUDIES IN A PRECLINICAL MODEL AND HUMAN SUBJECTS

Principal Investigator:
Gina Votta-Velis, MD, PhD
Department of Anesthesiology
University of Illinois Hospital and Health Sciences System

Co-Investigators:
Ajay Rana, PhD, Department of Surgery, / Cancer Center, UIC
Pier Giulianotti, MD, Department of Surgery, UIC
Antonio Gangemi, MD, Department of Surgery, UIC
P.J. Wheeler, MD, Department of Anesthesiology, UIC
Julie Golembiewski, Department of Aneshesiology, UIC

Key Personnel:
Alexandra Barabanova MS, Department of Anesthesiology, UIC

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SYNOPSIS

Primary Objective:

The primary objective of this study is to determine the effect of IV lidocaine infusion on Src tyrosine kinase activity in isolated CTCs as well as the number of CTCs during the perioperative period in patients undergoing robotic pancreatectomy for pancreatic cancer. We hypothesize that the lidocaine infusion will decrease Src tyrosine kinase activity in those CTCs that are released during surgery as well as the number of CTCs in the peripheral circulation.

Patient Population:

- Patients aged 18 and over
- Has histologically or cytologically confirmed adenocarcinoma of the pancreas that is at Stage IA-III and considered resectable
- American Society of Anesthesiology (ASA) status no > than III
- Have provided written informed consent

Study Design:

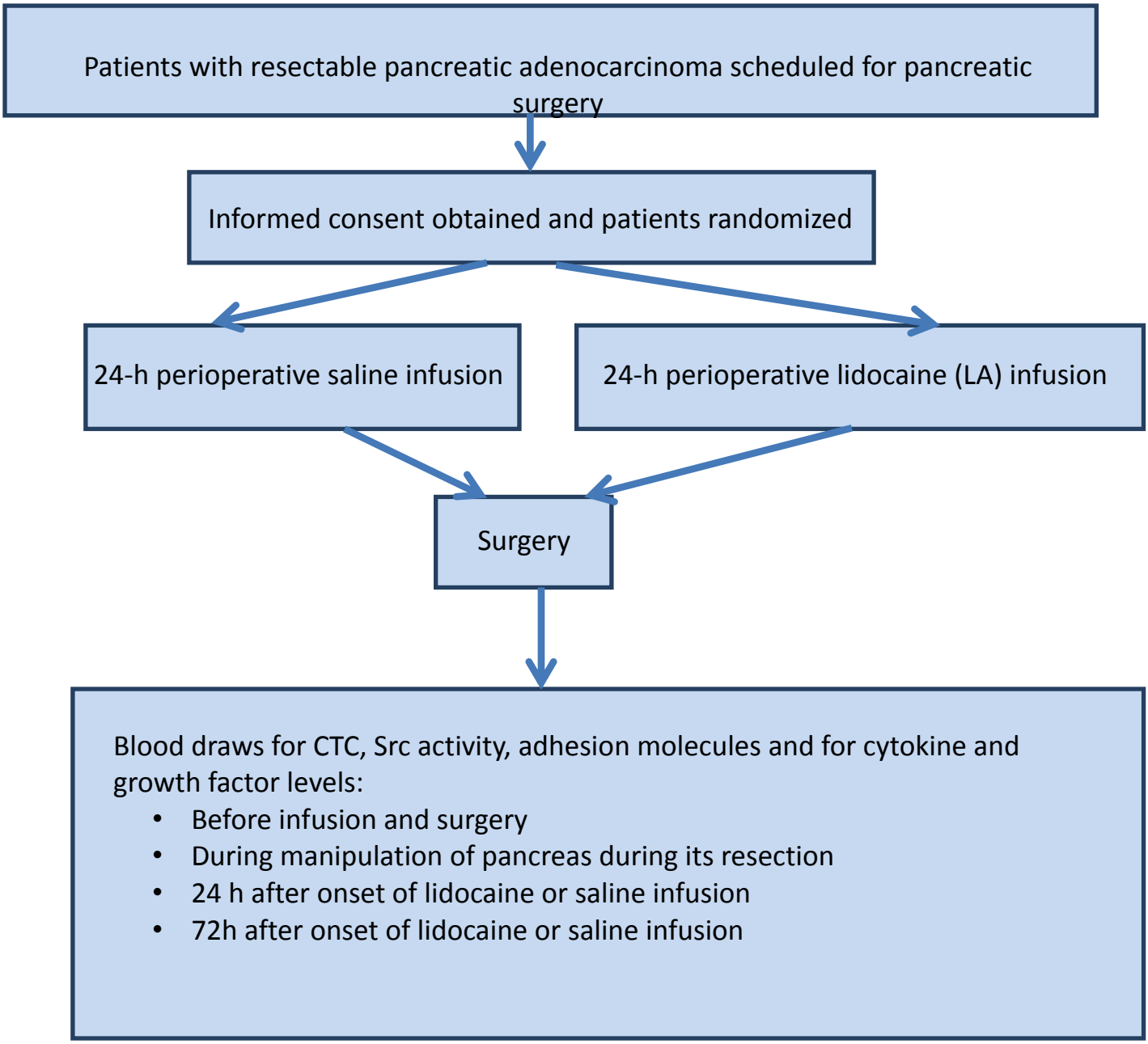
A prospective randomized controlled double blinded trial design will be used for the proposed study

Treatment Plan:

Patients undergoing robot-assisted laparoscopic pancreatic surgery for pancreatic cancer will be randomized (ratio 1:1) into two groups: one group will receive a 24-h normal saline infusion and the second group will receive a 24-h lidocaine infusion beginning shortly before administration of the anesthesia for the surgical procedure throughout the surgical period, and into the post-operative period for 24 hrs. Both groups will have their blood drawn for a total of 4 times: in the preoperative suite (baseline, before initiation of the infusion, during the surgery after the pancreas has been manipulated and 24 h, and 72 h after the initiation of the infusions.. Blood levels of CTCs and Src activity in the CTCs will be measured from the blood samples collected at these time points.

Other Src family kinases like Src A: Src, Yes, Fyn, and Fgr; Src B: Lck, Hck, Blk, and Lyn; Frk in its own subfamily will also be measured. Adhesion molecules, such as the intracellular adhesion molecule (ICAM-1) will be measured. Cytokines and chemokines arrays in the plasma will be measured from samples at baseline and 24h and 72 h after the initiation of the infusion. Study of gene expressions will be also performed in the 2 studied groups.

SCHEMA


Patients with resectable pancreatic adenocarcinoma scheduled for pancreatic surgery
Informed consent obtained and patients randomized
24-h perioperative saline infusion
24-h perioperative lidocaine (LA) infusion
Surgery
Blood draws for CTC, Src activity, adhesion molecules and for cytokine and growth factor levels:
• Before infusion and surgery
• During manipulation of pancreas during its resection
• 24 h after onset of lidocaine or saline infusion
• 72h after onset of lidocaine or saline infusion

ABBREVIATIONS

Abbreviation	Definition
AE	adverse event
ALT	alanine aminotransferase
ANC	absolute neutrophil count
ASA	American Society of Anesthesiologists
AST	aspartate aminotransferase
CMI	cell mediated immunity
CRF	case report form
CTC	circulating tumor cell
ECOG	Eastern Cooperative Oncology Group
EDTA	ethylene-diamine-tetra-acetic acid
ELISA	Enzyme-Linked Immunosorbent Assay
EMT	epithelial-mesenchymal transition
eNOS	endothelial nitric oxide synthase
h	hour
ICAM-1	intercellular adhesion molecule
IL	interleukin
IRB	institutional review board
IV	Intravenous
kg	kilogram
LA	local anesthetic agent
mg	milligram
min	minute
mL	milliliter
NK	natural killer
NKA	natural killer cell activity
NK-T	natural killer T
PAMAM	polyamidoamine

PDAC	pancreatic ductal adenocarcinoma
PEG	polyethylene glycol
PI3K	phosphatidylinositide 3-kinase
PMN	polymorphonuclear
Src	Src Tyrosine Kinase
TICU	Transplant Intensive Care Unit
TNFα	tumor necrosis factor-alpha
ULN	upper limit normal
US	United States
VEGF	vascular endothelial growth factor
wt	weight

1. BACKGROUND AND SIGNIFICANCE

1.1. Introduction

Pancreatic cancer, although not one of the most prevalent cancers, is highly aggressive and its detection is late in the process. It is estimated that it is the 8th cancer related cause of death in males and the 9th cause of death in females.[1] Pancreatic ductal adenocarcinoma (PDAC) accounts for >80% of all pancreatic neoplasms diagnosed.[2] The only curative treatment option for pancreatic cancer is surgical resection with approximately 15% of patients deemed suitable.[3] Unfortunately, the vast majority of patients diagnosed with pancreatic cancer often have distant metastases, and are not candidates for curative surgery. As a result, the prognosis for these patients is very poor with an overall 5-year survival rate of <5%.[4] Although this represents an overall survival benefit, in practical terms, the survival is less than a year. In recent years, there have been significant advances in our understanding of pancreatic tumor biology as well as a clearer understanding of the important role that the tumor microenvironment plays in tumor development and growth. Attempts at using targeted therapeutic agents in combination with standard chemotherapy have proved clinically meaningless with negligible improvements in survival. For these reasons, considerable challenges still remain in the treatment of patients with pancreatic cancer and as such, novel therapeutic approaches are urgently needed.

The standard treatment of solid malignant tumors including pancreatic is surgery. While a surgical approach can be effective in preventing long-term tumor recurrence, it may enhance the micrometastatic process in the immediate post-operative period. Several mechanisms have been proposed to explain the above observation.[5] These include suppression of cell mediated immunity (CMI),[6] an increase in proangiogenic factors,[7] and release of tumor cells into the circulation during and after surgery.[8]

1.2. Enhancement of the metastatic process via CMI suppression

CMI includes natural killer (NK) cell, natural killer T (NK-T) cell, dendritic cell, and macrophage cell functions. CMI affects the immune response to the circulating tumor cells (CTCs) and to the process of micrometastasis.[6] NK cells play a significant role in the intravascular elimination of CTCs.[9] Along with the surgical stress response, anesthetic agents also affect CMI. Inhalational anesthetics and opioids significantly reduce natural killer cell activity (NKA) and increase lung tumor metastasis.[10] It has also been shown that inhalational anesthetics upregulate the expression of hypoxia inducible factor-1α (HIF-1α) thereby enhancing cancer cell survival under hypoxic conditions.[11] Amongst opioids, morphine has been shown to have immunosuppressant properties and to increase survival of CTCs.[12]

1.3. Enhancement of the metastatic process via an increase in proangiogenic factors

Along with immunosuppressive properties, morphine has demonstrated proangiogenic properties. Angiogenesis (the formation of new blood vessels) plays an important role in the growth and metastasic potential of various cancers. It is mediated by degradation of the basement membrane, migration of endothelial cells towards an angiogenic stimulus, and proliferation of these cells. This results in the development of abnormal tumor vessels that are highly permeable to macromolecules and circulating inflammatory cells. One such molecule that is involved in these mechanisms is vascular endothelial growth factor (VEGF).[13] Morphine was shown to stimulate angiogenesis in a human breast tumor xenograft model in mice and promote tumor progression.[14] Our group has preliminary *in vitro* data demonstrating that the proangiogenic effect of morphine is attenuated by lidocaine.

1.4. Enhancement of the metastatic process via tumor cell release into the circulation

Tumor cell seeding during surgery plays a key role in tumor metastasis.[15] CTCs constitute the hematogenous route of metastasis and are the main cause of the development of distant metastatic sites.[16] Many studies have shown the presence of CTCs in the circulation in pancreatic cancer patients and the evidence seems to support their prognostic value in this type of cancer.

1.5. Src, Adhesion Molecules, and Inflammatory Cytokines Affect Tumor Metastasis

Once CTCs leave their primary site, they enter blood vessels and secrete VEGF which activates Src Tyrosine Kinase (Src) and causes endothelial barrier disruption, extravasation of CTCs, and formation of satellite lesions.[17] Src functions as a regulator of vascular permeability, cell adhesion, and migration.[18] Additionally, Src is involved in signaling epithelial-to-mesenchymal transformation (EMT) and extravasation of cancer cells,[19] processes that are necessary for solid tumor metastasis. Src phosphorylation of the intercellular adhesion molecule (ICAM-1) in endothelial cells facilitates neutrophil adhesion to the endothelium during an inflammatory response[20] and this is considered to be a key mechanism involved in the vascular hyperpermeability response.[21] Activation of Src family kinases, which is very common in a variety of human cancers, including pancreatic cancer,[22] (may occur via several mechanisms and is frequently a critical event in tumor progression. Because Src family kinases appear to play an important role in tumor proliferation, disruption of cell/cell contacts, migration, invasiveness and resistance to apoptosis, they are attractive targets for anticancer therapeutics.[23,24]

The metastatic process is also facilitated by adhesion molecules, such as ICAM-1, glycoproteins (CD11b,CD18), and others required for inflammation[25,26] ICAM-1 has been implicated in tumor invasion *in vitro,*[27] and in metastasis *in vivo*, hence it is thought to play a critical role in the malignant potential of various types of cancer. ICAM-1 expression in primary tumors and soluble ICAM-1 in serum of patients with malignancies is associated with reduced disease-free interval and survival,[28] and increased expression is associated with a more aggressive tumor phenotype.[29] For the above reasons, ICAM-1 can also be used as both a biomarker for tumor prognosis and a target for therapeutic interventions. Glycoproteins CD11b and CD18 expressed on human polymorphonuclear leukocytes (PMNs) are also implicated in PMN-facilitated tumor cell migration and extravasation.[30] Moreover, inflammatory cytokines in pancreatic cancer have been studied in depth, and they have been targeted by therapeutic agents as it has been postulated that they have a modulatory effect in tumor development. Increased levels of IL-6, IL-8 and IL-10, as well as low levels of IL-1RA correlate with poor clinical outcome.[31,32] In addition, the local and systemic release of inflammatory cytokines (e.g., TNFα, IL-1β, IL-6, IL-8, IL-1RA) and growth factors (such as VEGFc) from tissues injured during surgery[33,34] are thought to promote metastasis via Src activation in cancer cells [35].

1.6. Lidocaine – A Novel Therapeutic Intervention

Cancer dissemination is a multi-step process and the many cellular and molecular mechanisms involved are potential targets for therapeutic interventions. Traditional systemic therapy (i.e., chemotherapy and radiation therapy) is delayed for weeks after major surgery to allow wound healing and to avoid the risk of immune-suppression and postoperative infections.[34] This delay is associated with a worse outcome.[36] This may be because cellular and molecular events that are critical to the metastatic process (e.g., CTCs, Src, inflammatory cytokines) that are activated by the manipulation and removal of the tumor during surgery are not treated for weeks by systemic therapies that could have attenuated that activation.[37] Therefore a novel therapeutic intervention that does not have the toxicity of chemotherapy, and might attenuate the activation of the cellular and molecular events that are critical to the metastatic process during the perioperative period, presents a window of opportunity for cancer treatment that should not be missed.

In recent years, a numb er of retrospective studies have suggested that the perioperative use of regional anesthesia and local anesthetic agents (LAs) can reduce cancer-related mortality following surgical treatment of prostate[38,39] breast,[40] colorectal,[41,42] and ovarian[43] cancers as well as malignant melanoma.[44,45] Thus far, the beneficial effect of regional anesthesia and LAs on long-term outcome after cancer surgery has been attributed to the inhibition of the neuroendocrine stress response to surgery[6,34] and to the reduction in requirements of volatile anesthetics and opioids. Because volatile anesthetics have been implicated in suppressing CMI and morphine has been implicated in increasing proangiogenic factors, it is postulated that lidocaine may have a beneficial effect on long term outcome after cancer surgery because it reduces volatile anesthetic and opioid requirements.[46] It is also possible that lidocaine directly interferes with tumor metastasis by inhibiting inflammatory signaling. The actual mechanism by which regional anesthesia and local anesthetics might prove to be beneficial in cancer patients at the molecular level is currently under investigation by our group. It has been demonstrated that amide LAs have anti-inflammatory properties in addition to their anesthetic and analgesic effects. They exert their anti-inflammatory effects by intervening in several stages of the inflammatory pathway[47] and systemic lidocaine attenuates the stimulation of the inflammatory response induced by surgery. For example, lidocaine infusion attenuates plasma levels of IL-6, IL-8, IL-1RA, and complement C3a, as well as the expression of CD11b and P selectin.[48] The anti-inflammatory effects of lidocaine infusions are responsible for the faster recovery of bowel function after surgery.[48-52] *In vitro* studies have also demonstrated anti-inflammatory effects of LAs in models of acute vascular injury[53] as well as their antiproliferative and cytotoxic effects on cancer cells.[54,55]

There is increasing evidence that mechanisms similar to inflammatory processes play an important role in the development, growth, and metastasis of solid tumors.[56] The presence of inflammatory cells and inflammatory mediators in tumors, tissue remodeling, and angiogenesis is similar to the ones seen in chronic inflammatory responses that precede and constitute the hallmark of cancer-related inflammation.

As amide-linked LAs are known to have anti-inflammatory properties and as there is increasing evidence that inflammation and cancer share a connected pathway,[56] we hypothesized that LAs might attenuate the metastatic process of cancer cells, in a manner similar to that by which they attenuate inflammation (**see Figure 1**). In an effort to investigate the mechanism at the molecular level by which LAs attenuate cancer recurrence, we hypothesized that this effect may be due to their ability to inhibit Src tyrosine kinase activation. *In vitro* studies by our group demonstrated that amide local anesthetics (lidocaine and ropivacaine) at clinically-relevant concentrations dose-dependently inhibited TNFα-induced Src activation, ICAM-1 phosphorylation, and migration of human lung adenocarcinoma cells.[57] We have also shown that lidocaine at the clinically relevant concentration of 10 micromolar (μM) inhibited TNF-α-induced Src activation in a pancreatic cancer cell-line (Panc-1 cells)[76]

Furthermore we showed in lung microvascular endothelial cells that lidocaine and ropivacaine inhibit TNFα-induced inflammatory signaling by attenuating the recruitment of p85 subunit of PI3-kinase to TNF-receptor-1, thereby blocking subsequent Akt, eNOS, and Src activation and attenuating neutrophil adhesion and endothelial hyperpermeability.[58] This finding is significant because the PI3K/AKT pathway is also responsible for triggering a cascade of responses that enhance tumor progression, and molecules that block this pathway may increase cancer survival.[59]

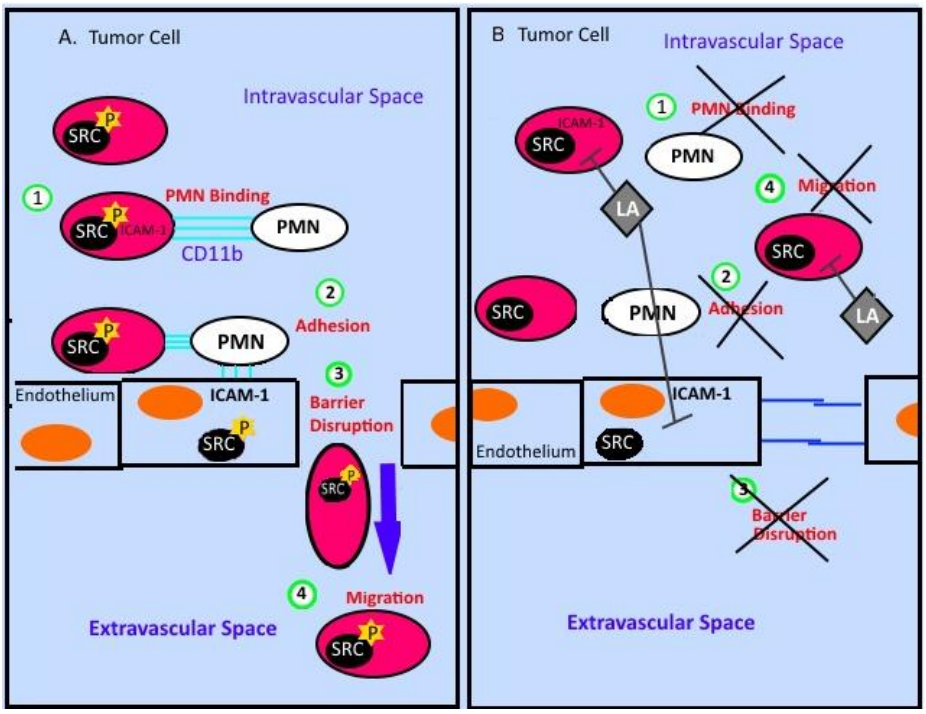


Figure 1. Proposed Mechanism of the Antimetastatic Effects of Lidocaine. Src tyrosine protein kinase activation (Src) in circulating tumor cells (CTCs) and in endothelial cells plays a role in tumor cell metastasis. A) Activation of Src by autophosphorylation (SrcP) leads to: increased PMN binding on CTCs (1) and adhesion to the vascular endothelium (2), stimulation of endothelial barrier disruption (3), enhanced migration and extravasation of the CTCs forming a new metastatic site (4). B) Lidocaine (LA) inhibition of Src kinase activation attenuates key steps involved in the extravasation of CTCs.

1.7. Innovation and Significance of the Proposed Study

Innovation: The notion that lidocaine, a therapeutic agent that has been used for its anesthetic and analgesic properties for more than 65 years, can have an entirely different application that could transform the perioperative anesthetic management of cancer patients, and favorably affect their outcome, is highly innovative.

Significance: Cellular and molecular events (e.g., CTCs) that are critical to the metastatic process may be significantly influenced perioperatively. Currently, the retrospective studies demonstrating the perioperative effect of regional anesthesia and LAs on cancer recurrence in patients undergoing cancer surgery provide us with weak evidence for intervention. Prospective RCTs are needed to establish the standard of care for the perioperative management of cancer patients. We have strong *in vitro* data to support the hypothesis that will be tested in this study. There are also preclinical studies in mice with very promising results demonstrating the effect of lidocaine infusions on tumor growth and metastasis. [77, 78] If we demonstrate reduced Src tyrosine kinase activity in CTCs as a result of our intervention, regional anesthesia and lidocaine infusion may acquire a distinct clinical application in the perioperative care of pancreatic cancer patients. Although the current study is a "Proof of Concept" study, if that concept is verified, it along with the retrospective studies would provide a more sound rationale for conducting a large, randomized controlled trial to assess the effect of lidocaine infusion on long term outcome.

2. OBJECTIVES

2.1. Primary Objective

To determine the effect of IV lidocaine infusion on Src tyrosine kinase activity in isolated CTCs as well as the number of CTCs during the perioperative period in patients undergoing robotic pancreatectomy for pancreatic cancer. We hypothesize that the lidocaine infusion will decrease Src tyrosine kinase activity in those CTCs that are released during surgery as well as the number of CTCs both in the peripheral

circulation and in the portal vein. Additional measures to asses other Src family kinases, adhesion molecules (ICAM-1) Cytokines, Chemokines and other proteins as above will be performed.

2.2. Secondary Objective

We will test the hypothesis that plasma and pancreatic tissue samples obtained from patients who have received lidocaine infusions, will demonstrate a reduced expression of gene signals linked to metastasis relative to the samples from the patients who did not receive lidocaine infusions

3. OVERALL DESIGN AND STUDY PLAN

A prospective randomized placebo-controlled double blinded trial design will be used for the proposed study. The study will be blinded, i.e., the surgeon performing the pancreatic surgery will be unaware whether the patient is receiving normal saline or lidocaine.

4. SELECTION OF PATIENTS

Inclusion criteria

1. Male or female ≥ 18 years of age
2. Has histologically or cytologically confirmed adenocarcinoma of the pancreas that is at Stage IA-III and considered resectable
3. Has measurable disease, defined as at least 1 tumor that fulfills the criteria for a target lesion according to RECIST 1.1
4. Has an Eastern Cooperative Oncology Group (ECOG) performance status of 0, 1, or 2
5. Has been off chemotherapy for > or = 2 weeks
6. Has adequate hepatic function defined as total bilirubin ≤ 2 mg/dL, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) ≤ 3.0 × upper limit of normal (ULN)
7. Has adequate renal function defined as serum creatinine ≤ 2.5 × ULN
8. Has adequate bone marrow function defined as a hemoglobin ≥ 10 g/dL, absolute neutrophil count (ANC) ≥ 1.5×10^9/L, and platelet count ≥ 100×10^9/L
9. Must be willing and able to comply with study visits and procedures
10. Has read, understood and signed the informed consent form (ICF) approved by the Independent Review Board/Independent Ethics Committee (IRB/IEC)
11. Prior systemic treatments for metastatic disease are permitted, including targeted therapies, biologic response modifiers, chemotherapy, hormonal therapy, or investigational therapy.

Exclusion Criteria

1. Has a diagnosis of unresectable pancreatic adenocarcinoma
2. Has American Society of Anesthesiologists (ASA) physical status > 3
3. Has hypersensitivity or allergy to amide-linked local anesthetics
4. Has a second or third degree heart block
5. Has severe sinoatrial block
6. Is currently being treated with any of the following class I antiarrhytmic drugs; quinidine, flecainide, disopyramide, or procainamide
7. Has been treated with amiodarone HCl in the past
8. Has Adams-Stoke syndrome
9. Has Wolff-Parkinson-White syndrome
10. Has a history of blood clots, pulmonary embolism, or deep vein thrombosis unless controlled by anticoagulant treatment
11. Has a known history of human immunodeficiency virus (HIV) positivity or untreated and uncontrolled hepatitis B or C

12. Has any clinically significant infection, i.e., any acute viral, bacterial, or fungal infection that requires specific treatment (anti-infective treatment has to be completed ≥ 7 days prior to study entry)
13. Pregnant or breastfeeding. Confirmation that the subject is not pregnant must be established by a negative serum beta-human chorionic gonadotropin (beta-hCG) pregnancy test result obtained during screening. Pregnancy testing is not required for post-menopausal or surgically sterilized women.
14. Other severe acute or chronic medical or psychiatric conditions, or laboratory abnormality that may increase the risk associated with study participation or may interfere with the interpretation of study results and, in the judgment of the investigator, would make the patient inappropriate for enrollment in this study
15. Has any condition that, in the opinion of the investigator, might jeopardize the safety of the patient or interfere with protocol compliance
16. Has any mental or medical condition that prevents the patient from giving informed consent or participating in the trial

Further Exclusions and Removals from the Study:If a patient signs consent and is registered to the study, and is later found not able to undergo the surgery for whatever reason, the patient will be removed from study and treated at the physician's discretion. The patient will be considered a screen/baseline failure and be replaced. The reason for removal from study will be clearly indicated in the Principal Investigator's data management system. In the unlikely event of patients exhibiting bradycardia or arrhythmias in the TICU, the infusion will be terminated, the blind will be broken, and the patient will receive appropriate treatment. At that point the patient will be considered off study. After the lidocaine infusion is terminated depending on the patient's condition he or she will remain there or will be transferred to a hospital room on a regular floor. The subject's pain in the TICU will be treated with hydromorphone (IV) and Tylenol (IV). Subjects will be released from the medical center when they are fit for discharge, as determined by their medical team.

In some cases, after Dr. Giulianotti begins the robot-assisted laparoscopic pancreatic surgery (see Section 6.1, below), it may become necessary depending on the nature of the tumor and its surrounding environment to switch from the robot-assisted laparoscopic surgery to open pancreatic surgery. It is estimated that approximately 5% of the cases will involve a conversion. If this occurs, the subject at this point will be terminated from the study, and the infusion pump will be turned off, while the open pancreatic surgery proceeds

5. REGISTRATION

In order to increase participation, we will be sending an recuirement email to campus listserv and posting recruitment flier on campus and in the hospitals and academic centers throught the US to recruite our participants. Registration into the study will occur after the patient has signed the patient consent and eligibility is confirmed, but before any treatment has been administered. Eligibility will be determined based on the patient's preoperative evaluation done at the UIHHSS Anesthesia Preoperative Care Clinic, which includes a physical, history, blood work, and assessment of cardiac and other organ functioning in order to clear the patient for surgery according to the American Society of Anesthesiology Guidelines (see Section 4, Inclusion and Exclusion criteria). To be eligible for registration to this study, the patient must meet each criteria listed on the eligibility checklist based on the eligibility assessment documented in the patient's medical record. A copy of the eligibility checklist (see Appendix A) is maintained by Principal Investigator and can be found for each

individual subject in a folder that contains the subject's signed informed consent form. All folders are stored in Dr. Votta-Velis' office (1740 West Taylor Street, Suite 3200 W, UIH) in a locked filing cabinet.

Assignment to either receiving the lidocaine infusion or to the saline infusion will be determined at registration, by the investigational drug pharmacist, using standard randomization tables.

6. TREATMENT PLAN

6.1. Experimental setting and procedures

The study will take place in a general operating room at UI-Hospital and Health Sciences System, and postoperatively in the Transplant Intensive Care Unit. The surgical procedure that will be performed is robot-assisted laparoscopic pancreatic surgery.[60] A standardized general anesthetic regimen will be provided to all subjects. General anesthesia will be performed using propofol as an induction agent and the inhalational agent sevoflurane for maintenance. Muscle relaxation will be achieved with vecuronium or rocuronium. Pain will be managed intraoperatively with intravenous (IV) administration of opioid hydromorphone. The IV bolus and infusions of lidocaine to those patients assigned to the lidocaine group will be started in the operating room approximately just before induction of general anesthesia, and will continue until 24 h later. The group receiving the lidocaine infusion will first be administered a 1.5 mg/kg loading infusion over 5 minutes followed by a 1.5 mg/kg/h infusion for 24 h (see below for dosing rationale). Lidocaine used for this study will be kept in the OR pharmacy. IV lidocaine is currently approved by the FDA for the treatment of ventricular arrhythmias and is routinely used for the manangement of peri-operative pain. We will be administering the IV lidocaine according our hospital guidelines for the management of peri-operative pain. However, since we are investigating the potential anti-metastatic effect of lidocaine infusion, the use of lidocaine will be considered investigational. Therefore, the cost of lidocaine infusion for the treatment group and of saline (placebo) infusion for the control group will be paid by the principal investigator's research funds . The group receiving the saline infusion will be administered an equivalent volume of saline infused over 5 min followed by a saline infusion at the same flow rate as that used in the lidocaine group for 24 h. The length of the robot-assisted laparoscopic pancreatic surgery is estimated to be approximately 8 h.[61] After the surgery, the subject will be transported to the Transplant Intensive Care Unit (TICU). Cardiac functioning will be continuously monitored at the TICU. In the unlikely event of patients exhibiting bradycardia or arrhythmias in the TICU, the infusion will be terminated, the blind will be broken, and the patient will receive appropriate treatment. At that point the patient will be considered off study. After the lidocaine infusion is terminated depending on the patient's condition he or she will remain there or will be transferred to a hospital room on a regular floor. The subject's pain in the TICU will be treated with hydromorphone (IV) and Tylenol (IV). Subjects will be released from the medical center when they are fit for discharge, as determined by their medical team.

Blood will be drawn for research purposes at four time points: in the preoperative area (peripheral vein), during the surgery when the pancreas is being manipulated (both from the portal vein and peripherally), , and 24 h, and 72 h after the start of the infusion (peripheral vein) in order to evaluate the time course of lidocaine concentrations. All specimens will be collected into BD Vacutainer™ tubes containing either Lithium Heparin or EDTA and transported to Dr. Rana's lab for processing (see below). Labeling of the tubes is described in Section 9, Subject Confidentiality. During the surgery, after the tumor is removed, it will be immediately taken to the pathology laboratory. Tissue samples will be divided and placed in liquid nitrogen or freshly made paraformaldehyde (4%) and after labeling (described in Section 9, Subject Confidentiality), will be transferred

to Dr.Rana's lab for analysis of Src activity in tumor tissue by Western blot and immunostaining/confocal microscopy.

Rationale and description of procedure of drawing blood from the portal vein for collection of CTCs:
Portal vein blood draw. We are drawing blood from the portal vein because there is a greater probability of detecting more CTCs in the portal vein than in peripheral blood. A recent study evaluated the feasibility and safety of sampling portal venous blood via endoscopic ultrasound (EUS) to count portal venous circulating tumor cells (CTCs), compared with paired peripheral CTCs, in patients with pancreaticobiliary cancers (PBCs).[75] There were no complications from portal vein blood acquisition. CTCs were detected in portal vein samples from all 18 patients (100%) vs peripheral blood samples from only 4 patients (22.2%). Patients with confirmed PBCs had a mean of 118.4 ± 36.8 CTCs/7.5 mL portal vein blood, compared with a mean of 0.8 ± 0.4 CTCs/7.5 mL peripheral blood ($P < .01$). Nine patients with nonmetastatic, resectable, or borderline-resectable PBCs had a mean of 83.2 CTCs/7.5 mL portal vein blood (median, 62.0 CTCs/7.5 mL portal vein blood).

The blood sampling will be done using a robotic approach with the da Vinci Robot.

The da Vinci Robot has 4 arms (1 camera and 3 instruments)

The blood sample will be drawn from the portal vein indirectly through cannulation of the inferior mesenteric vein (IMV) which is a very small vein inside the body. The IMV is a blood vessel that drains blood from the large intestine and terminates at the splenic vein which subsequently joins the superior mesenteric vein and form together the portal vein (see anatomy Figure 2). Drawing blood from the IMV is a relatively safe procedure as in this way we avoid direct puncture of the portal vein, which could lead to rare complications such as thrombosis and bleeding.

Surgeon #1 (Dr. Giulianotti sitting in the Robotic console) with the robotic scissors will be performing a small cut of 1-2mm in the inferior mesenteric vein wall.
Surgeon #2 who is assisting with the procedure at the operating room table will be introducing a small 4 French (1.32 mm outer diameter) catheter to the one of the robotic trocars that are used to provide access to the peritoneal cavity after pulling out the robotic instruments. (No extra hole in the abdominal wall). The catheter is made out of silicone.
After making a tiny nick in the inferior mesenteric vein, surgeon #1 will grasp the tip of the catheter that has been inserted through the trocar in the abdominal cavity, with the robotic forceps, and place it inside the lumen of the inferior mesenteric vein.
A 20cc Syringe will be attached to the distal extremity of the 4 fr. Catheter and 12 cc of blood will be drawn by Dr. Giulianotti and his surgical team. The blood will be transferred immediately to the appropriate Vacutainer Heparin tubes (6ml/tube) will be stored at room temperature and will be transferred to Dr. Rana's Lab by Alexandra Barabanova (Research Assistant).
Dr. Giulianotti will pull out the catheter from the inferior mesenteric vein and will eliminate the risk of bleeding by placing a suture tie in the vein.
Since there are many collateral veins there will not be any consequences for the patient.
The blood drawing from the portal vein system will add one minute to the surgical procedure. This process will be performed only once in the controlled environment of the operating room under direct visualization.

Blood drawing from the portal vein system is performed for diagnostic purposes for follow up for lesions in the liver and, is a standard procedure for hormone producing tumors. Several studies in the '80s described the

use of percutaneous transhepatic sampling of blood in the portal vein system (TPVS) for the localization of hormone secreting tumors and to help differentiating tumors from diffuse disease.
See References below:

***REF #1*:** Gastrointestinal / Pancreatic hormone Concentrations in the portal venous system of nine patients with organic hyperinsulinism
Benjamin Glaser et al.
Metabolism 30 (10) : October 1981.

Also cannulation of the inferior mesenteric vein is a procedure that is performed by Dr. Giulianotti for islet cell transplantation. (Figure 3)
REF#2: Robot- assisted pancreatoduodenectomy with preservation of the vascular supply for autologous islet cell isolation and transplantation: a case report.
Giulianotti et al.
Journal of Medical Case Reports. 2012, 6:74

Finally islet cell transplantation through cannulation of the portal vein is performed by interventional radiology.
REF #3: Pancreatic Islet Cell Transplantation: An update for Interventional Radiologists.
Gaba et al; J Vasc Interv Radiol 2012; 23: 583-594

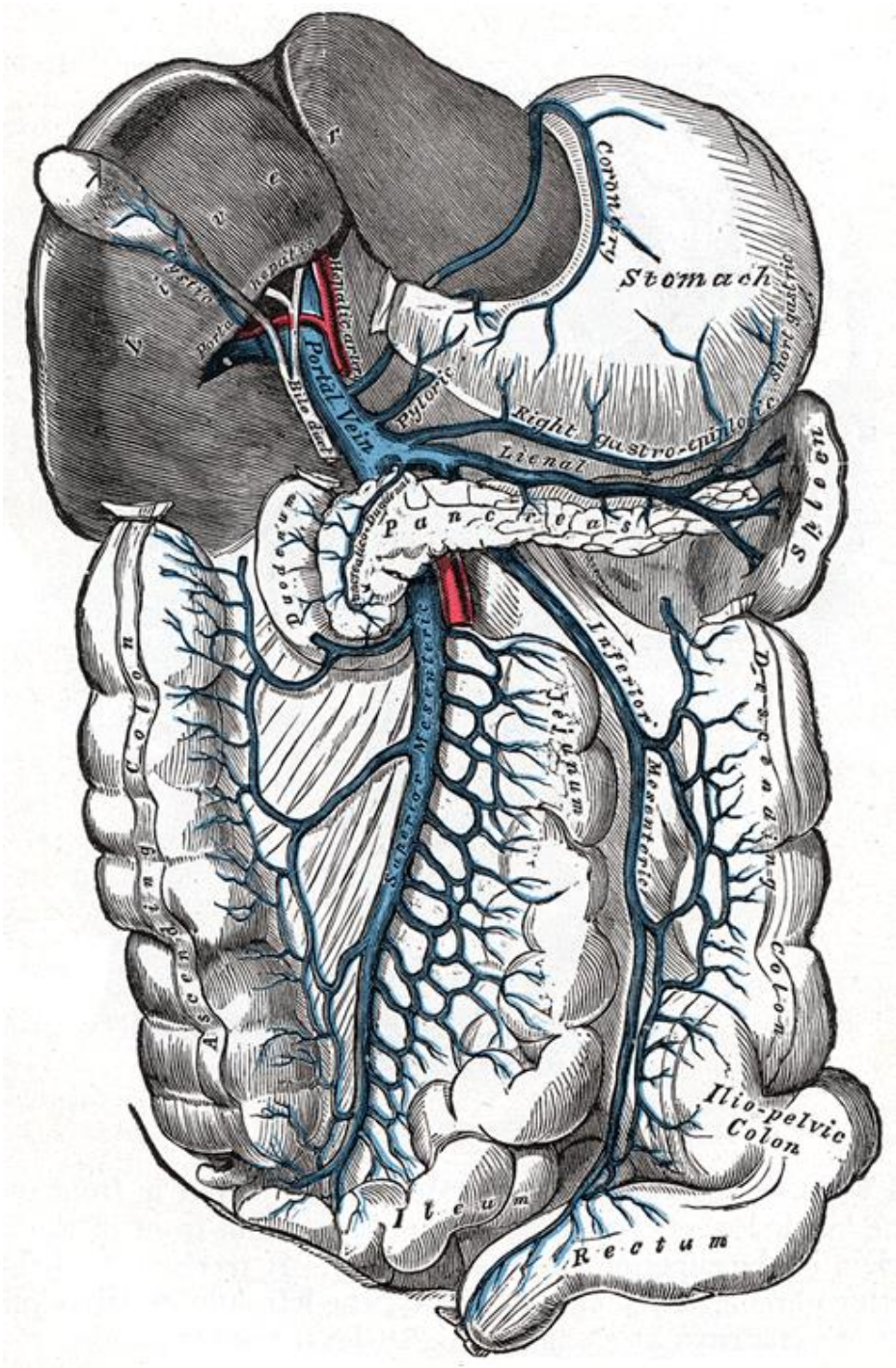


Figure 2. Abdominal Anatomy

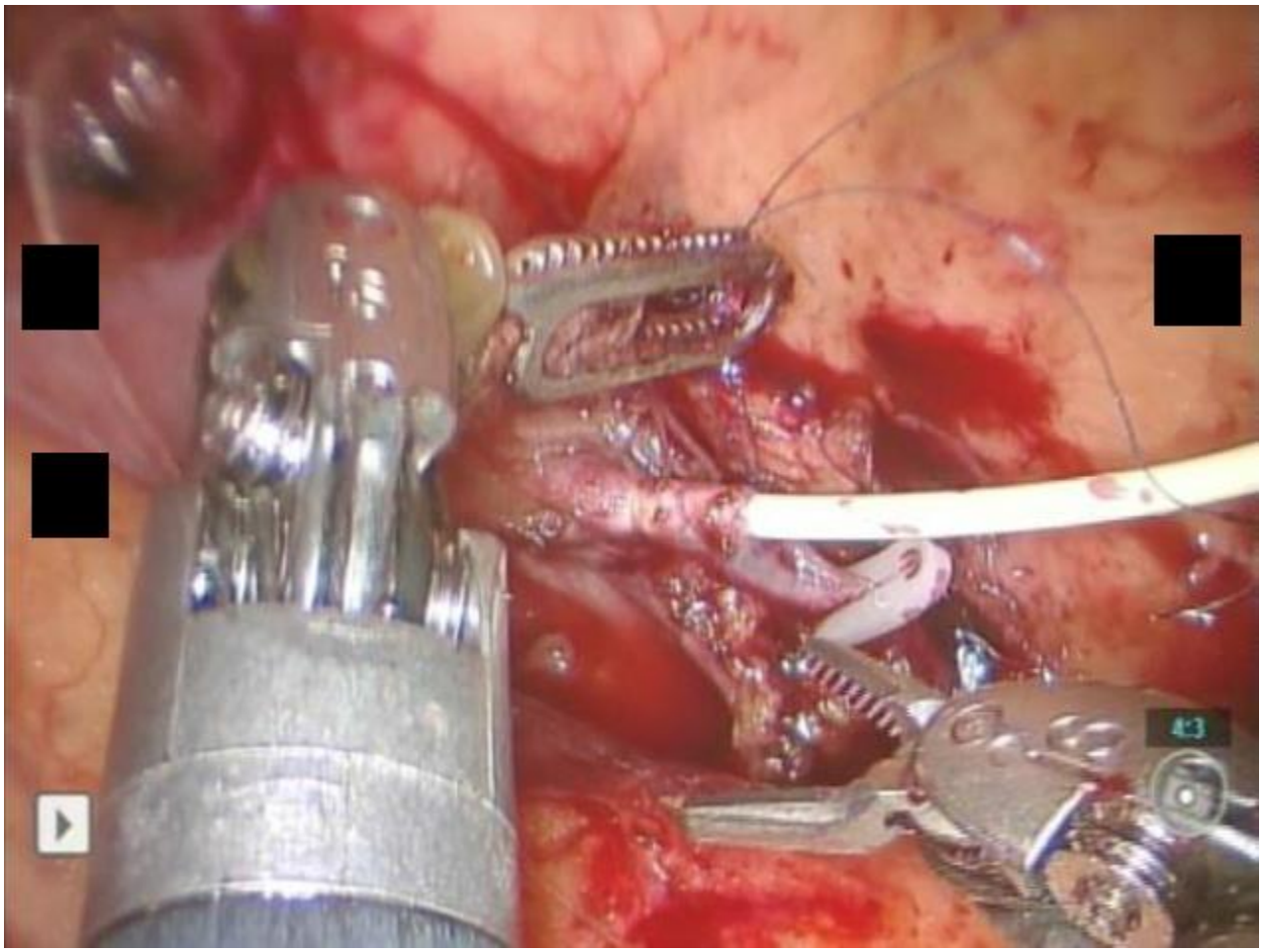


Figure 3. Ligation of the inferior mesenteric vein. The inferior mesenteric vein was identified, dissected and distally ligated. A canula was inserted for infusion of the pancreatic digest for autologous islet transplantation. Cannulation of the inferior mesenteric vein is a procedure that is performed by Dr. Giulianotti for islet cell transplantation.

See REF below.
REF: Robot- assisted pancreatoduodenectomy with preservation of the vascular supply for autologous islet cell isolation and transplantation: a case report. *Giulianotti et al. Journal of Medical Case Reports. 2012, 6:74*

6.2. Rationale for lidocaine dose to be administered in the study

The proposed lidocaine infusion (a 1.5 mg/kg loading infusion over 5 minutes followed by a 1.5 mg/kg/hr infusion for 24 hr) was simulated for an 80 kg patient with the SAAM II software system (SAAM Institute, Seattle, WA) implemented on a Windows^{TM}-based PC using the multicompartmental pharmacokinetic parameters of Kuipers et al.[62] Plasma lidocaine concentrations are predicted to rapidly plateau at approximately 2 μg/mL (Figure 4), or 8.53 μM, which is consistent with the concentration we have demonstrated to produce a significant decrease in TNF-α- induced Src phosphorylation of 73% after coincubation of cells with TNF-α, 10 μM lidocaine.[57] In some patients iv lidocaine infusions were given at higher concentrations and durations than that to be used in the proposed study – no adverse incidents were

reported.[64] Lidocaine infusions have analgesic and antihyperalgesic properties and are currently used commonly world wide as part of the enhanced recovery after surgery (ERAS) protocols.[63 74]

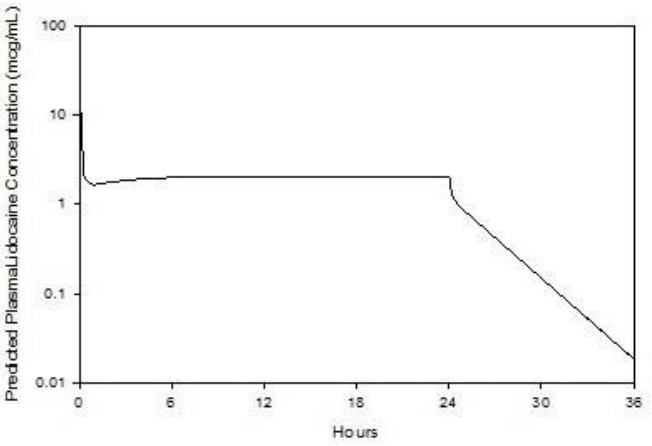


Fig. 4 Simulated plasma lidocaine concentrations during and after a 1.5 mg/kg loading infusion over 5 min followed by a 1.5 mg/kg/h infusion for 24 h in an 80 kg patient.

7. STUDY ENDPOINTS
Study endpoints

The table below outlines when and how key outcome measures will be collected.

	Bio specimen Outcome Measure					
	Src[a]		**CTCs[b]**		**Cytokines/ other proteins[c]**	
Sample Collection Time	lidocaine	saline	lidocaine	Saline	lidocaine	Saline
Preop	x	x	x	x	x	x
During surgery[d,e]	x	x	x	x		
24-h post-lidocaine infusion	x	x	x	x	x	x
72-h post-lidocaine infusion	x	x	x	x	x	x

a+b - whole blood drawn in 2 Vacutainer tubes (6 mL per tube) containing lithium heparin
c - 4 mL whole blood drawn in EDTA-containing plasma test tube
d - specifically after the pancreas has been manipulated
e – blood will be drawn both peripherally and through a portal vein

7.1. Experimental Protocol
7.1.1. Methods for purification of CTCs and Src kinase activity.

All CTC enumerations will be performed in Prof. Ajay Rana Lab at UIC. Using magnetic separations of micro beads. CTCs will be isolated from whole blood of patients using Tumor cell isolation Kit, human (Miltenyi Biotec) based on the lineage markers including CD45, CD31 and Ter119. Cells will be characterized using flow cytometry. Src and ICAM activation state will be determined by immunohistochemical staining and Western Blot analysis in the Rana lab as described for CTCs isolated from KPC mice using human specific antibodies.

7.1.2. Cytokines and Other Proteins.

Serum and plasma will be collected from human subjects for the estimation of cytokines. The levels of 13 human inflammatory cytokines/chemokines, including IL-1β, IFN-α, IFN-γ, TNF-α, MCP-1 (CCL2), IL-6, IL-8 (CXCL8), IL-10, IL-12p70, IL-17A, IL-18, IL-23, and IL-33 will be studied using LEGENDplex™ Human Inflammation Panel (BioLegend). For estimation of VEGFc enzyme linked immunosorbent assay (ELISA) kit will be used. All assays will be performed per the manufacturer's protocols. Concentrations of each target protein in serum or plasma will be calculated from the standard curve.

7.1.3. Single-cell Microfluidics-based RT-PCR analysis.

To get broad idea about the gene expressions associated with Src activity single cell RNA sequencing will be performed. Single-cell microfluidics-based RNA sequencing will be carried out at Core Genomic Facility (CGF) at UIC. In brief, microfluidics-based RT-PCR analysis will be performed using CellsDirectTM one-step qRT-PCR kit (Invitrogen) and a microfluidics device, BioMark HD MX/HX system (Fluidigm, Inc). Single CTCs in PBS/lysis buffer will be thawed, mixed well and spin down before lysed at 75°C for 10 min. To reduce contamination, genomic DNA will be degraded in an 18 μg reaction volume using DNase I (5 units) with 1X DNase I buffer at RT for 5 min. PCR primers of selected genes for expression profiling will be selected from the Primer Bank database. These primers will be divided into two panels to fit BioMark 48x48 chips. Reverse transcription, preamplification, and PCR amplification will be carried out according to the protocol of single-cell gene expression (Fluidigm). Target genes will be amplified using BioMark HB MX/HX system with 1X SsoFast EvaGreen super mix with low ROX (Bio-Rad) and 1X DNA binding dye sample loading reagent (Fluidigm). In each chip assay, universal RNA (200 pg) from human normal tissues and no template control (NTC) served as positive and negative controls.

7.1.4. In vitro silencing of specific genes using siRNA.

Following single cell RNA sequencing, most relevant gene/s in reference to metastasis will be studied to understand further mechanisms. The gene of interest will be silenced in CTCs using siRNA for that specific gene/s. siRNA for the scrambled control will be used for experimental control. The transfection of siRNA will be carried out using Amaxa® Nucleofector® II System for delivery of siRNA. Knock down will be confirmed by reverse transcriptase PCR or Western blots.

7.1.5. Opioid Consumption

Study personnel will collect data pertaining to type, dose, frequency, and route of administration of opioids during surgery and for 72 hours postoperatively. As a tertiary outcome measure, total opioid amount used in both groups will be compared.

8. DRUG FORMULATION

8.1. Lidocaine

8.1.1. Other Names

Xylocaine®, Lignocaine

8.1.2. Classification

Local anesthetic from the amide group

8.1.3. How Supplied

Lidocaine Hydrochloride and 5% Dextrose Injection, USP, 2 grams/500 mL (4 mg/mL), VIAFLEX Plus Plastic Container. NDC: 0338-0409-03 Baxter 2B0973

It’s a sterile intravenous solution,

8.1.4. Availability

Lidocaine is commercially available.

8.1.5. Description

Sterile, nonpyrogenic, aqueous solution

8.1.6. Storage, Handling, and Accountability

Stored at room temperature with a pH of 4.0 (range: 3.0 – 7.0).

8.1.7. Administration

The IV bolus and infusions of lidocaine to those patients assigned to the lidocaine group will be started in the operating room approximately 10 minutes before induction of general anesthesia, and will continue until 24 h later. The group receiving the lidocaine infusion will first be administered a 1.5 mg/kg loading infusion over 5 minutes followed by a 1.5 mg/kg/h infusion for 24.0 h.

8.1.8. Risks

The potential risks to subjects are minimal, given the lidocaine dosage we are employing[48,51], and the setting in which the drug is being given. The setting in which the drug will be given are in an anesthesiology operating area and an intensive care unit at the UIC Medical Center containing equipment for monitoring vital signs and for resuscitation. In our setting, drug interventions are supervised by licensed medical professionals. Side effects from the administered Lidocaine at 1.5 mg/kg/hr for 24 hrs are highly unlikely. In the rare event they occur, they may include feeling lightheaded, shaking, low blood pressure, drowsiness, confusion, weakness, blurry or double vision, and dizziness. Less likely side effects (serious adverse events) are lidocaine toxicity (this occurs with doses much higher [5-10 times] than those that will be tested in this study) that includes cardiovascular effects including hypotension, bradycardia (slower hear rate) which may lead to cardiac arrest, and cardiopulmonary arrest ,nervous system effects including seizures, gastrointestinal effects including nausea and vomiting, hypersensitivity (allergic reactions) including anaphylaxis, adult respiratory distress syndrome (ARDS), and cardiovascular collapse and that have occasionally resulted in death, psychiatric reactions including transient (not permanent) psychoses, and hematologic effects limited to rare reports of significant increases in methemoglobin levels.

9. SUBJECT CONFIDENTIALITY

After enrolling a subject into the study, pertinent information regarding the subject will be entered into a password-protected Microsoft Excel database on a password-protected server in the Department of Anesthesiology, by Dr, Votta-Velis or designated members of her research team. In the database, each patient will have a unique identifier (coded subject ID number) assigned by Dr. Votta-Velis. The unique identifier will be placed into a Master ID file linking the identifier to the patient's EMR. Only Dr. Votta-Velis (study PI) will have access to the Master ID file linking a subject's unique identifier (coded ID number) to their UI Health medical chart and their electronic medical record (EMR) with subject identifiers. The Master ID file will be on the password-protected server in the Department of Anesthesiology. Information entered into the password-protected Microsoft Excel database will include demographic, clinical, and laboratory variables. Demographic variables will include subject sex, birth date, race/ethnicity, and height and weight. Clinical variables will include current diagnoses, date and duration of surgery, opioid consumption during and after surgery, histology, and tumor grade. Laboratory values will include Src kinases activity, adhesion molecules, CTC number, and plasma levels of cytokines and other proteins, a single cell RNA sequencing and silencing of specific genes in isolated CTCs.

The tubes containing the blood samples collected before, during, and after surgery, and the pancreatic tissue sample obtained from the Department of Pathology (see section 6.1), for each subject in this study will be assigned the same unique identifier (coded subject ID number) by Dr. Votta-Velis that is used for the Microsoft Excel database. Any personal identifying information associated with the biospecimens (electronic

medical record number, patient name) will be removed by Dr. Votta-Velis, and the unique subject identifier along with the Principal Investigator's last name and UIC IRB number will be put in its place. Only Dr. Votta-Velis (study PI) and Alex Barabanova (Study coordinator) will have access to the Master ID file linking the specimen unique identifier (coded subject ID number) to the subject's UI Health medical chart and their electronic medical record (EMR).

10STUDY DATA COLLECTION AND MONITORING

10.1. Data Management

This study will report clinical data utilizing study specific case report forms. Key study personnel are trained on the use of case report forms and will comply with protocol specific instructions for data collection.

Patient demographics, adverse events and other information required for PRC and IRB annual reporting will be maintained by the Principal Investigator or designated members of her research staff using Microsoft Excel databases on password-protected secure servers in the Department of Anesthesiology.

10.2. Case Report Forms

Participant data will be collected using protocol specific case report forms (CRFs). The CRFs will be approved by the study's Principal Investigator and the study biostatistician prior to release for use. The Study Coordinator or designee will be responsible for registering the patient into the Principal Investigator's password-protected data management system at time of study entry, completing CRFs based on the patient specific calendar, and updating the patient record until end of required study participation.

10.3. Data Safety Monitoring Plan (DSMP)

Lidocaine will be administered to 50% of the subjects in this protocol. Given the safety record of this FDA-approved drug, and the setting in which it will be administered, the risk of serious toxicities should be low. We do not anticipate the occurrence of any serious adverse events. One of the potential adverse event associated with this study involves loss of confidentiality/breach of privacy. We will report any privacy breach to the IRB as part of the annual continuing review. If a breach of patient privacy/confidentiality occurs that the study PI views as greater than a minor occurrence, the IRB will be notified of such at the time of discovery, prior to the annual continuing review, and a mitigation plan will be developed in consultation with the IRB.

Adverse Events Documentation and Reporting:

Serious adverse events may be spontaneously reported by the patient and/or in response to an open question from study personnel or revealed by observation, physical examination, or other diagnostic procedures. Serious adverse events occurring from lidocaine infusion must be documented. These serious adverse events were listed in section 8.1.8. (Risks) as "less likely" and are extremely rare, and include lidocaine toxicity (this occurs with doses much higher [5 to 10 times] than those that will be tested in this study) that include cardiovascular effects including hypotension, bradycardia (slower hear rate) which may lead to cardiac arrest,

and cardiopulmonary arrest, nervous system effects including seizures, gastrointestinal effects including nausea and vomiting, hypersensitivity (allergic reactions) including anaphylaxis, adult respiratory distress syndrome (ARDS), and cardiovascular collapse and that have occasionally resulted in death, psychiatric reactions including transient (not permanent) psychoses, and hematologic effects limited to rare reports of significant increases in methemoglobin levels. Serious adverse events attributed to a study-related procedure which occur prior to the initiation of study treatment must be documented as well.

Relationship to the study drug (lidocaine) for each serious adverse event will be determined by the investigator or sub-investigator by responding yes or no to the question: Is there a reasonable possibility that the serious adverse event is associated with the study drug. If the answer is "yes" the IRB must be notified within 5 working days using the UIC IRB Prompt Report Form.

10.4. Monitoring

The investigator will permit study-related monitoring, audits, and inspections by the local IRB, government regulatory bodies, and University of Illinois compliance groups. The investigator will make available all study related documents (e.g. source documents, regulatory documents, data collection instruments, study data, etc.). The investigator will ensure that all applicable study-related facilities (e.g. pharmacy) will be available for trial related monitoring, audits, or regulatory inspections.

10.5 Record Retention

The investigator will retain study records including source data, copies of case report form, consent forms, HIPAA authorizations, and all study correspondence in a secured facility for at least 6 years after the study file is closed with the IRB.

11 STATISTICAL CONSIDERATIONS

Data will be expressed as the means +/- SD. Statistical analysis will be conducted to assess differences between the two groups with regards to Src activity, CTC number, and cytokine/other proteins compared to pre-treatment levels. A mixed-design two-way repeated-measures analysis of variance (ANOVA) with Bonferroni *post hoc* testing will be used (between groups factor: lidocaine vs. normal saline; within groups factor: blood collection time points). Parameter estimates, 90% confidence intervals and Wald tests will be conducted and results will be expressed as a percentage of basal level. We will also perform multivariate ANOVA analysis that examines the group differences among the vector changes of all dependent variables (Src activity, CTC number, and cytokine/other proteins). $P < 0.05$ will be considered significant.(STATA 14)

12. Power and Sample Size

We performed the following sample size calculations to ensure adequate power for the study. According to previous results obtained *in vitro*, application of ropivacaine decreased Src activity by 30%.[56] In the present patient study, we consider a 30% reduction in Src activation to be clinically significant. A power analysis, which was performed by setting the level of significance at 5% and the power at 90%, determined a sample size of 20 patients for each of the two groups as necessary to achieve significance with within-group standard deviation of 20%. Assuming that there will be subjects who do not have evaluable data for the reasons cited in

section 5.3., we project that we will need to recruit 42-46 subjects. The power analyses were performed using the software PASS (Kaysville Utah 2008).

13. CONDUCT OF THE STUDY

13.1. Good Clinical Practice

The study will be conducted in accordance with the appropriate regulatory requirement(s). Essential clinical documents will be maintained to demonstrate the validity of the study and the integrity of the data collected. Master files should be established at the beginning of the study, maintained for the duration of the study and retained according to the appropriate regulations. All the investigators will have current CITI training required by the IRB.

13.2. Ethical Considerations

The study will be conducted in accordance with ethical principles founded in the Declaration of Helsinki. The IRB will review all appropriate study documentation in order to safeguard the rights, safety and well-being of the patients. The study will only be conducted at sites where IRB approval has been obtained. The protocol, consent, written information given to the patients, safety updates, progress reports, and any revisions to these documents will be provided to the IRB by the Investigator.

14. REFERENCES

1. Jemal A, Bray F, Center MM, Ferlay J, Ward E, Forman D. Global cancer statistics. CA Cancer J Clin. 2011;61:69-90.
2. Seufferlein T, Bachet JB, Van Cutsem E, Rougier P; ESMO Guidelines Working Group. Pancreatic adenocarcinoma: ESMO-ESDO Clinical Practice Guidelines for diagnosis, treatment and follow-up. Ann Oncol. 2012;23 Suppl 7:vii33-40.
3. Loos M, Kleeff J, Friess H, Büchler MW Surgical treatment of pancreatic cancer.. Ann N Y Acad Sci. 2008;1138:169-80.
4. Li D, Xie K, Wolff R, Abbruzzese JL. Pancreatic cancer. Lancet. 2004;363:1049-57.
5. Shakhar G, Ben-Eliyahu S. Potential prophylactic measures against postoperative immunosuppression: Could they reduce recurrence rates in oncological patients? Ann Surg Oncol. 2003;10:972-92.
6. Ben-Eliyahu S, Page G, Yirmiya R, Shakhar G. Evidence that stress and surgical interventions promote tumor development by suspending natural killer cell activity. Int J Cancer 1999;80:880-8.
7. Holmgren L, O' Reilly MS, Folkman J. Dormancy of micrometastases, balanced proliferation and apoptosis in the presence of angiogenesis suppression. Nat Med. 1995;1:149-53.
8. Weitz J, Herfarth C. Surgical strategies and minimal residual disease detection. Semin Surg Oncol. 2001;20: 329-33.
9. Zamai L, Ponti C, Mirandola P, Gobbi G, Papa S, Galeotti L, Cocco L, Vitale M. NK cells and Cancer. J Immunol. 2007;178:4011-6.
10. Melamed R, Bar-Yosef S, Shakhar G, Shakhar K, Ben-Eliyahu S. Suppression of natural killer cell activity and promotion of tumor metastasis by ketamine, thiopental, and halothane, but not by propofol: mediating mechanisms and prophylactic measures. Anesth Analg. 2003;97:1331-9.
11. Ma D, Lim T, Xu J, Tang H, Wan Y, Zhao H, Hossain M, Maxwell PH, Maze M. Xenon preconditioning protects against renal ischemic reperfusion injury via HIF-1 alpha activation. J Am Soc Nephrol. 2009;20:713-720.
12. Roy S, Wang J, Kelschenbach J, Koodie L, Martin J. Modulation of immune function by morphine. implications for susceptibility to infection. J. Neuroimmune Pharmacol. 2006:1:77-89.
13. Carmelier P. Mechanisms of angiogenesis and arteriogenesis. Nature Med. 2000*;*6:389-95.
14. Gupta K, Kshirsagar S, Chang L, Schwartz R, Law PY, Yee D, Hebbel RP. Morphine stimulates angiogenesis by activating proangiogenic and survival-promoting signaling and promotes breast tumor growth. Cancer Res. 2002;62:4491-8.
15. Pachmann K. Tumor cell seeding during surgery – possible contribution to metastasis formations. Cancers (Basel) 2011;3:2540-53.
16. Eschwege P, Dumas F, Blanchet P, Le Maire V, Benoit G, Jardin A, Lacour B, Loric S. Haematogenous dissemination of prostatic epithelial cells during radical prostatectomy. Lancet 1995;346:1528-30.
17. Weiss S, Cui J, Barnes L, Cheresh D. Endothelial barrier disruption by VEGF-mediated Src activity potentiates tumor cell extravasation and metastasis. J Cell Biol. 2004;167:223-9.
18. Hu G, Minshall RD. Regulation of transendothelial permeability by Src kinase. Microvasc Res. 2009;77:21-5.
19. Guarino M. Src signaling in cancer invasion. J Cell Physiol. 2010;223:14-26.
20. Liu G, Vogel SM, Gao X, Javaid K, Hu G, Danilov SM, Malik AB, Minshall RD. Src phosphorylation of endothelial cell surface intercellular adhesion molecule-1 mediates neutrophil adhesion and contributes to the mechanism of lung inflammation. Arterioscler Thromb Vasc Biol. 2011;31:1342-50.
21. Hu G, Vogel SM, Schwartz DE, Malik AB, Minshall RD. Intercellular adhesion molecule-1-dependent neutrophil adhesion to endothelial cells induces caveolae-mediated pulmonary vascular hyperpermeability. Circ Res. 2008;102:e120-31.

22. Nagaraj NS, Smith JJJ, Revetta F, Washington MK, Merchant NB. Targeted inhibition of SRC kinase signaling attenuates pancreatic tumorigenesis. Mol Cancer Ther. 2010;9:2322-32.
23. Summy JM, Gallick GE. Src family kinases in tumor progression and metastasis. Cancer Metastasis Rev. 2003;22:337–58.
24. Wong, H. H., & Lemoine, N. R. (2009). Pancreatic cancer: molecular pathogenesis and new therapeutic targets. *Nat Rev Gastroenterol Hepatol, 6*(7), 412-422. doi:10.1038/nrgastro.2009.89
25. Lin YC, Shun CT, Wu MS, Chen CC. A novel anticancer effect of thalidomide: inhibition of intercellular adhesion molecule-1-mediated cell invasion and metastasis through suppression of nuclear factor-kappaB. Clin Cancer Res. 2006;12:7165-73.
26. Roland CL, Dineen SP, Toombs JE, Carbon JG, Smith CW, Brekken RA, Barnett CC Jr. Tumor-derived intercellular adhesion molecule-1 mediates tumor-associated leukocyte infiltration in orthotopic pancreatic xenografts. Exp Biol Med(Maywood) 2010;235:263-70.
27. Liu X, Chen Q, Yan J, Wang Y, Zhu C, Chen C, Zhao X, Xu M, et al. MiRNA-296-3p-ICAM-1 axis promotes metastasis of prostate cancer by possible enhancing survival of natural killer cell-resistant circulating tumour cells. Cell Death Dis. 2013; 4:e928.
28. Kageshita T, Yoshii A, Kimura T, Kuriya N, Ono T, Tsujisaki M, Imai K, Ferrone S. Clinical relevance of ICAM-1 expression in primary lesions and serum of patients with malignant melanoma. Cancer Res. 1993; 53:4927-32.
29. Roland CL, Harken AH, Sarr MG, Barnett CC Jr. ICAM-1 expression determines malignant potential of cancer. Surgery 2007;141:705-7.
30. Wu QD, Wang JH, Condron C, Bouchier-Hayes D, Redmond HP. Human neutrophils facilitate tumor cell transendothelial migration. Am J Physiol Cell Physiol. 2001;280:C814-22.
31. Miron N, Miron MM, Milea VG, Cristea V. Proinflammatory cytokines: an insight into pancreatic oncogenesis, Roum. Arch. Microbiol. Immunol. 2010;69:183–189.
32. Smirne C, Bellone G, Mauri FA, Tonel E, Carbone A, Buffolino A, Dughera L, Robecchi A, Pirisi M, Emanuelli G, Cytokine expression profile in human pancreatic carcinoma cells and in surgical specimens: implications for survival. Cancer Immunol. Immunother. 2006;55:684–698.
33. Hofer SO, Molema G, Hermens RA, Wanebo HJ, Reichner JS, Hoekstra HJ. The effect of surgical wounding on tumor development. Eur J Surg Oncol. 1999;25:231-43.
34. Desborough JP: The stress response to trauma and surgery; Br J Anaesth. 2000;85:109-17.
35. Liu ST, Pham H, Pandol SJ, Ptasznik A. Src as the link between inflammation and cancer. Front Physiol. 2014;4:416.
36. Chhatwani L, Cabebe E, Wakelee HA. Adjuvant treatment of resected lung cancer. Proc Am Thorac Soc. 2009;6:194-200.
37. Biagi JJ, Raphael MJ, Mackillop WJ, Kong W, King WD, Booth CM. Association between time to initiation of adjuvant chemotherapy and survival in colorectal cancer: a systematic review and meta-analysis. JAMA. 2011;305:2335-42.
38. Biki B, Mascha E, Moriarty DC, Fitzpatrick JM, Sessler DI, Buggy DJ. Anesthetic technique for radical prostatectomy surgery affects cancer recurrence: a retrospective analysis. Anesthesiology 2008;109:180-7.
39. Wuethrich PY, Hsu Schmitz SF, Kessler TM, Thalmann GN, Studer UE, Stueber F, Burkhard FC. Potential influence of the anesthetic technique used during open radical prostatectomy on prostate cancer related outcome: a retrospective study. Anesthesiology 2010;113:570-6.
40. Exadaktylos AK, Buggy DJ, Moriarty DC, Mascha E, Sessler DI. Can anesthetic technique for primary breast cancer surgery affect recurrence or metastasis? Anesthesiology 2006;105:660-4.
41. Gottschalk A, Ford JG, Regelin CC, You J, Mascha EJ, Sessler DI, Durieux ME, Nemergut EC. Association between epidural analgesia and cancer recurrence after colorectal cancer surgery. Anesthesiology 2010;113:27–34.

42. Cummings KC 3rd, Xu F, Cummings LC, Cooper GS. A comparison of epidural analgesia and traditional pain man-agement effects on survival and cancer recurrence after colectomy: a population-based study. Anesthesiology 2012;116:797-806.
43. de Oliveira GS Jr, Ahmad S, Schink JC, Singh DK, Fitzgerald PC, McCarthy RJ. Intraoperative neuraxial anesthesia but not postoperative neuraxial analgesia is associated with increased relapse-free survival in ovarian cancer patients after primary cytoreductive surgery. Reg Anesth Pain Med. 2011;36:271-7.
44. Schlagenhauff B, Ellwanger U, Breuninger H, Stroebel W, Rassner G, Garbe C. Prognostic impact of the type of anaesthesia used during the excision of primary cutaneous melanoma. Melanoma Res. 2000;10:165-9.
45. Gottschalk A, Brodner G, Van Aken HK, Ellger B, Althaus S, Schulze HJ. Can regional anaesthesia for lymph-node dissection improve the prognosis in malignant melanoma? Br J Anaesth. 2012;109:253-9.
46. De Oliveira GS, Jr , Fitzgerald P, Streicher LF, Marcus RJ, McCarthy RJ. Systemic lidocaine to improve postoperative quality of recovery after ambulatory laparoscopic surgery. Anesth Analg. 2012;115:262-7.
47. Hollmann, M, Durieux M. Local anesthetics and the inflammatory response. A new therapeutic indication? Anesthesiology 2000 93:858-75.
48. Herroeder S, Pecher S, Schonherr ME, Kaulitz G, Hahnenkamp K, Friess H, Bottiger BW, Bauer H, *et al*. Systemic lidocaine shortens length of hospital stay after colorectal surgery: a double-blinded, randomized, placebo-controlled trial. Ann Surg. 2007;246:192-200.
49. Kuo CP, Jao SW, Chen KM, Wong CS, Yeh CC, Sheen MJ, Wu CT. Comparison of the effects of thoracic epidural analgesia and i.v. infusion with lidocaine on cytokine response, postoperative pain and bowel function in patients undergoing colonic surgery. Br J Anaesth 2006;97:640-6.
50. Groudine SB, Fisher HA, Kaufman RP, Jr, Patel MK, Wilkins LJ, Mehta SA, Lumb PD. Intravenous lidocaine speeds the return of bowel function, decreases postoperative pain, and shortens hospital stay in patients undergoing radical retropubic prostatectomy. Anesth Analg. 1998; 86: 235-9.
51. Kaba A, Laurent SR, Detroz BJ, Sessler DI, Durieux ME, Lamy ML, Joris JL. Intravenous lidocaine infusion facilitates acute rehabilitation after laparoscopic colectomy. Anesthesiology 2007;106:11–8.
52. Swenson B, Gottschalk A. Wells L, Rowlingson J, Thompson P, Barclay M, Sawyer R, Friel M, Foley E, Durieux M. Intravenous lidocaine is as effective as epidural bupivacaine in reducing ileus, duration , hospital stay, and pain after open colon resection. Reg Anesth Pain Med, 2010;35:370-6.
53. Blumenthal S, Borgeat A, Pasch T, Reyes L, Booy C, Lambert M, Schimmer RC, Beck-Schimmer B. Ropivacaine decreases inflammation in experimental endotoxin-induced lung injury. Anesthesiology 2006;104:961-9.
54. Martinsson T. Ropivacaine inhibits serum-induced proliferation of colon adenocarcinoma cells in vitro. J Pharmacol Exp Ther. 1999;288: 660-4.
55. Lucchinetti E, Awad AE, Rahman M, Feng J, Lou PH, Zhang L, Ionescu L, Lemieux H, *et al.* Antiproliferative effects of local anesthetics on mesenchymal stem cells: potential implications for tumor spreading and wound healing. Anesthesiology 2012;116: 841–56.
56. Mantovani A, Allavena P, Sica A, Balkwill F. Cancer-related inflammation. Nature 2008; 454:436–44.
57. Piegeler T, Votta-Velis G, Liu G., Place AT, Schwarz DE, Beck-Schimmer B, Minshall RD, Borgeat A. Anti-metastatic potential of amide-linked local anesthetics: Inhibition of lung adenocarcinoma cell migration and inflammatory Src signaling independent of sodium channel blockade. Anesthesiology 2012;117;548-59.
58. Piegeler T, Votta-Velis G, Bakhshi MS, Mao SS, Carnegie GK, Bonini MG, Schwartz DE, Borgeat A, Beck-Schimmer B, Minshall RD. Endothelial barrier protection by local anesthetics: Ropivacaine and lidocaine block tumor necrosis factor-α-induced endothelial cell Src activation. Anesthesiology 2014;120:1414-28.

59. Vivanco I, Sawyers CL. The Phosphatidylinositol 3-Kinase-AKT pathway in human cancer. Nat Rev Cancer 2002;2:489-501.
60. Fernandes E, Giulianotti PC. Robotic-assisted pancreatic surgery. J Hepatobiliary Pacreatr Sci 2013, April 16. PMID: 23588851.
61. Buchs NC, Addeo P, Bianco FM, Ayloo S, Benedetti E, Giulianotti PC. Robotic versus open pancreaticoduodenectomy: a comparative study at a single institution. World J Surg. 201;35:2739-46.
62. Kuipers JA1, Boer F, de Roode A, Olofsen E, Bovill JG, Burm AG. Modeling population pharmacokinetics of lidocaine: Should cardiac output be included as a patient factor? Anesthesiology. 2001;94:566-73.
63. Roger Chou, Debra B. Gordon, Oscar A. de Leon-Casasola, Jack M. Rosenberg, Stephen Bickler, Tim Brennan, et al. Management of Postoperative Pain: A Clinical Practice Guideline From the American Pain Society, the American Society of Regional Anesthesia and Pain Medicine, and the American Society of Anesthesiologists' Committee on Regional Anesthesia, Executive Committee, and Administrative Council. The Journal of Pain; Volume 17, Issue 2, Pages 131–157

64. Williams D, Stark R. Intravenous Lidocaine infusion for the treatment of chronic daily headache with substantial medication overuse. Cephalalgia. 2003;;23(:963-71.
65. Osta WA, Chen Y, Mikhitarian K, Mitas M, Salem M, Hannun YA, Cole DJ, Gillanders WE. EpCAM is overexpressed in breast cancer and is a potential target for breast cancer gene therapy. Cancer Res. 2004;64:5818-24.
66. Sainsbury JR, Farndon JR, Needham GK, Malcolm AJ, Harris AL. Epidermal-growth-factor receptor status as predictor of early recurrence of and death from breast cancer. Lancet 1987;1:1398-402.
67. Kindler HL, Friberg G, Singh DA, Locker G, Nattam S, Kozloff M, Taber DA, Karrison T, Dachman A, Stadler WM, Vokes EE. Phase II trial of bevacizumab plus gemcitabine in patients with advanced pancreatic cancer. J Clin Oncol. 23:8033-40.
68. Yin T, Wei H, Leng Z, Yang Z, Gou S, Wu H, Zhao G, Hu X, Wang C. Bmi-1 promotes the chemoresistance, invasion and tumorigenesis of pancreatic cancer cells. Chemotherapy. 2011;57:488-96.
69. Wu PF, Lu ZP, Cai BB, Tian L, Zou C, Jiang KR, Miao Y. Role of CXCL12/CXCR4 signaling axis in pancreatic cancer. Chin Med J (Engl). 2013;126:3371-4.
70. Li L, Hao X, Qin J, Tang W, He F, Smith A, Zhang M, Simeone DM, Qiao XT, Chen ZN, Lawrence TS, Xu L. Antibody against CD44s inhibits pancreatic tumor initiation and postradiation recurrence in mice. Gastroenterology. 2014 Apr;146(4):1108-18.
71. Armstrong AJ, Marengo MS, Oltean S, Kemeny G, Bitting RL, Turnbull JD, Herold CI, Marcom PK, *et al.* Circulating tumor cells from patients with advanced prostate and breast cancer display both epithelial and mesenchymal markers. Mol Cancer Res. 2011;9:997-1007.
72. Myung JH, Gajjar KA, Chen J, Molokie RE, Hong S. Differential detection of tumor cells using a combination of cell rolling, multivalent binding, and multiple antibodies. Anal Chem. 2014;86:6088-94.
73. Place AT, Chen Z, Bakhshi FR, Liu G, O'Bryan JP, Minshall RD. Cooperative role of caveolin-1 and C-terminal Src kinase binding protein in C-terminal Src kinase-mediated negative regulation of c-Src. Mol Pharmacol. 2011;80:665-72.
74. Dunn LK, Durieux ME. Perioperative use of intravenous lidocaine. Anesthesiology. 2017; 126(4):729-737.
75. Catenacci DV, Chapman CG, Xu P, Koons A, Konda VJ, Siddiqui UD, Waxman I. Acquisition of Portal Venous Circulating Tumor Cells From Patients With Pancreaticobiliary Cancers by Endoscopic Ultrasound. Gastroenterology. 2015 Dec;149(7):1794-1803.e4.

76. Francesco Vetri, Sin-jung Park, Ja Hye Myung, Alain Borgeat, Arkadiusz Z. Dudek, Seungpyo Hong, and Gina Votta-Velis.Evaluation of the Effects of Lidocaine on Src Kinase Activity in Vitro in a Pancreatic Cancer Cell Line. *Oral presentation at the Annual meeting of the American Society of Anesthesiology (ASA), Chicago IL, October 2016.*

77. Wei Xing MD, PhD., Dong-Tai Chen, M.D. Jia-Hao Pan, M.D. et al. Lidocaine Induces Apoptosis and Supresses Tumor Growth in Human Hepatocellular Carcinoma Cells In Vitro and in a Xenograft Model In Vivo. Anesthesiology 2017; V 126:868-881

78. Johnson MZ, Crowley PD, Foley AG, Xue C, Connolly C, Gallagher HC, Buggy DJ. Effect of perioperative i.v. lidocaine on metastasis after sevoflurane or ketamine-xylazine anaesthesia for breast tumour resection in the 4T1 murine model. Br J Anaesth (in press)

Appendix A – Eligibility Checklist
LIDOCAINE INFUSIONS IN PANCREATIC CANCER: TRANSLATIONAL STUDIES IN A PRECLINICAL MODEL AND HUMAN SUBJECTS
Eligibility Checklist Page 1 of 2
Patient initials ☐☐☐☐ Patient ID☐☐☐
1st 2 initials of first name + 1st 2 initials of last name

INCLUSION CRITERIA
A "NO" response to any of the following disqualifies the patient from study entry.

		Yes	No
1.	Male or female ≥ 18 years of age	☐	☐
2.	Has histologically or cytologically confirmed adenocarcinoma of the pancreas that is at Stage IA-III and considered resectable	☐	☐
3.	Has measurable disease, defined as at least 1 tumor that fulfills the criteria for a target lesion according to RECIST 1.1	☐	☐
4.	Has an Eastern Cooperative Oncology Group (ECOG) performance status of 0, 1, or 2	☐	☐
5.	Has been off chemotherapy for > or = 2 weeks	☐	☐
6.	Has adequate hepatic function defined as total bilirubin < 2 mg/dL, aspartate aminotransferase (AST) and alanine aminotransferase (ALT) < 3.0 × upper limit of normal (ULN)	☐	☐
7.	Has adequate renal function defined as serum creatinine < 2.5 × ULN	☐	☐
8.	Has adequate bone marrow function defined as a hemoglobin ≥ 10 g/dL, absolute neutrophil count (ANC) ≥ 1.5 × 109/L, and platelet count ≥ 100 × 109/L	☐	☐
9.	Must be willing and able to comply with study visits and procedures	☐	☐
10	Has read, understood and signed the informed consent form (ICF) approved by the Independent Review Board/Independent Ethics Committee (IRB/IEC)	☐	☐
11	Prior systemic treatments for metastatic disease are permitted, including targeted therapies, biologic response modifiers, chemotherapy, hormonal therapy, or investigational therapy	☐	☐

EXCLUSION CRITERIA

A "YES" response to any of the following disqualifies the patient from study entry.

		Yes	No
12.	Has a diagnosis of unresectable pancreatic adenocarcinoma	☐	☐
13.	Has American Society of Anesthesiologists (ASA) physical status > 3	☐	☐
14.	Has hypersensitivity or allergy to amide-linked local anesthetics	☐	☐
15.	Has a second or third degree heart block	☐	☐
16.	Has severe sinoatrial block	☐	☐
17.	Is currently being treated with any of the following class I antiarrhytmic drugs; quinidine, flecainide, disopyramide, or procainamide	☐	☐
18.	Has been treated with amiodarone HCl in the past	☐	☐
19.	Has Adams-Stoke syndrome	☐	☐
20.	Has Wolff-Parkinson-White syndrome	☐	☐
21.	Has a history of blood clots, pulmonary embolism, or deep vein thrombosis unless controlled by anticoagulant treatment	☐	☐
22.	Has a known history of human immunodeficiency virus (HIV) positivity or untreated and uncontrolled hepatitis B or C	☐	☐
23.	Has any clinically significant infection, i.e., any acute viral, bacterial, or fungal infection that requires specific treatment (anti-infective treatment has to be completed ≥ 7 days prior to study entry)	☐	☐
24.	Pregnant or breastfeeding. Confirmation that the subject is not pregnant must be established by a negative serum beta-human chorionic gonadotropin (beta-hCG) pregnancy test result obtained during screening. Pregnancy testing is not required for post-menopausal or surgically sterilized women.	☐	☐
25.	Other severe acute or chronic medical or psychiatric conditions, or laboratory abnormality that may increase the risk associated with study participation or may interfere with the interpretation of study results and, in the judgment of the investigator, would make the patient inappropriate for enrollment in this study	☐	☐
26.	Has any condition that, in the opinion of the investigator, might jeopardize the safety of the patient or interfere with protocol compliance	☐	☐
27.	Has any mental or medical condition that prevents the patient from giving informed consent or participating in the trial	☐	☐

Date consent form signed: _____

Having obtained consent and reviewed each of the inclusion/exclusion criteria, I verify that this patient is:
☐ Eligible ☐ Ineligible Date registered _____

_____
Signature of person verifying eligibility